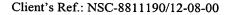
Client's Ref.: NSC-8811190/12-08-00

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WHAT IS CLAIMED IS:

- 1 1. An integrated plasmid comprising a biotin synthase
- 2 gene, an assistant DNA sequence for the integration of said
- 3 plasmid into a host genome, a promoter sequence, and a
- 4 selection marker.
- 1 2. The integrated plasmid as claimed in claim 1,
- 2 wherein the biotin synthase gene is derived from
- 3 Saccharomyces cerevisae or Candida utilis.
- 3. The integrated plasmid as claimed in claim 2,
- 2 wherein the biotin synthase gene of Candida utilis comprises
- 3 the nucleotide sequence of SEQ ID NO: 1.
- 4. The integrated plasmid as claimed in claim 1,
- 5 wherein the assistant DNA sequence is a Candida utilis
- 6 fragment selected from the group consisting of NsiI-BamHI
- 7 18s rDNA, URA3 DNA, and HIS3 DNA.
- 5. The integrated plasmid as claimed in claim 1,
- 2 wherein the selection marker is a cycloheximide-resistant
- 3 gene.
- 1 6. The integrated plasmid as claimed in claim 1,
- 2 wherein the promoter sequence is selected from the group
- 3 consisting of pL41 promoter of Candida utilis and pADH1
- 4 promoter of Saccharomyces cerevisae.

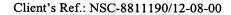


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genome.

```
1
              7. The integrated plasmid as claimed in claim 1,
     2
        wherein the integrated plasmid is selected from the group
        consisting of:
     3
     4
              (a) pMCC21 (having the configuration of restriction
     5
        sites in FIG. 6);
              (b) pMCC31S (having the configuration of restriction
     6
     7
        sites in FIG. 8);
     8
              (c) pMCC32H (having the configuration of restriction
     9
        sites in FIG. 9);
              (d) pMCC33U (having the configuration of restriction
   10
   11
        sites in FIG. 10);
Mr. Hom Gen. and Home man
   12
              (e) pMCC35U (having the configuration of restriction
   13.
        sites in FIG. 11);
   14
              (f) pMCC36H (having the configuration of restriction
15
        sites in FIG. 12); and
              (g) pMCC38S (having the configuration of restriction
   16
   17
        sites in FIG. 13).
             98. A method for preparing a yeast with high biotin-
        productivity, \comprising the steps of:
    2
    3
             constructing an integrated plasmid comprising a biotin
    4
        synthase gene, ah assistant DNA sequence for the integration
        of said plasmid into a host genome, a promoter sequence, and
    5
    6
        a selection marker)
             linearizing said integrated plasmid;
    8
             transforming said linearized integrated plasmid into a
    9
        yeast; and
   10
             recombining the biotin synthase gene with the yeast
```



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- 9. The method as claimed in claim 8, wherein the biotin
- 2 synthase gene is derived from Saccharomyces cerevisae or
- 3 Candida utilis.
- 1 10. The method as claimed in claim 9, wherein the
- 2 biotin synthase gene of Candida utilis comprises the
- 3 nucleotide sequence of SEQ ID NO: 1.
- 1 11. The method as claimed in claim 8, wherein the
- 2 assistant DNA sequence is a Candida utilis fragment selected
- 3 from the group consisting of NsiI-BamHI 18s rDNA, URA3 DNA,
- 4 and HIS3 DNA.
- 1 12. The method as claimed in claim 8, wherein the
- 2 selection marker is a cycloheximide-resistant gene.
- 1 13. The method as claimed in claim 8, wherein the
- 2 promoter sequence is selected from the group consisting of
- 3 pL41 promoter of Candida utilis and pADH1 promoter of
- 4 Saccharomyces cerevisae.
- 1 14. The method as claimed in claim 8, wherein the
- 2 prepared yeast with high biotin-productivity is useful as
- 3 feed additives, food additives, or cosmetics.
- 1 15. A method for producing biotin, comprising:
- 2 providing the yeast with high biotin-productivity of
- 3 claim 8; and
- 4 culturing said yeast in a nutrient medium, and
- 5 recovering biotin from the culture broth.



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- 1 16. The method as claimed in claim 15, wherein the
- 2 recovered biotin is useful as feed additives, food additives,
- 3 or cosmetics.